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Frank C Eisenschenk
Frank C. Eisenschenk, Ph.D., Patent Attorney

REQUEST FOR CERTIFICATE OF
CORRECTION UNDER 37 CFR 1.322
Docket No. DAS-103XC1

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicants : Frank H. Gaertner, Stacey Finstad Lee, Robert Shutter
Issued : March 4, 2008
Patent No. : 7,338,794
For : Amended Recombinant Cells for the Production and Delivery of Gamma Interferon as an Antiviral Agent, Adjuvant and Vaccine Accelerant

Mail Stop Certificate of Corrections Branch
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

## REQUEST FOR CERTIFICATE OF CORRECTION UNDER 37 CFR 1.322 (OFFICE MISTAKE)

Sir:

A Certificate of Correction for the above-identified patent has been prepared and is attached hereto.

In the left-hand column below is the column and line number where errors occurred in the patent. In the right-hand column is the page and line number in the application where the correct information appears.

| **Patent Reads**: | **Application Reads**: |
|---|---|
| Column 4, line 8: | Page 5, paragraph [0021], line 12: |
| “2α/GROP” | --2α/GROβ-- |
| Column 11, lines 17-18: | Page 15, paragraph [0047], line 6: |
| “1’-16, 11-18 ” | --11-16, 11-18-- |

| | |
|---|---|
| Column 23, line 17: | Page 31, paragraph [0085], line 6: |
| “MVP(T” | --MVP®-- |
| Column 24, line 48: | Page 33, paragraph [0090], line 7: |
| “ $\geqq 10^{10.6}$” | --$\geq 10^{10.6}$-- |
| Column 25, line 20: | Page 34, paragraph [0093], line 7: |
| “400%1” | --400μ1-- |
| Column 28, lines 13-14: | Page 38, line 2: |
| “bovine tracheitis” | --bovine rhino tracheitis-- |
| Column 32, Table 8, MCP-1/MCAF Reference, Function column: | Amendment Under 37 C.F.R. §1.111 dated January 29, 2007 (page 5): |
| “aguments neutrophil” | --augments neutrophil-- |
| Column 32, Table 8, MIP-1β Reference, Function column: | Amendment Under 37 C.F.R. §1.111 dated January 29, 2007 (page 5): |
| “moncytes, densritic” | --monocytes, dendritic-- |
| Column 37, lines 58-59: | Amendment Under 37 C.F.R. §1.111 dated January 29, 2007, claim 2: |
| “gene encodes” | --gene that encodes-- |
| Column 38, lines 60-61: | Amendment Under 37 C.F.R. §1.111 dated January 29, 2007, claim 2: |
| “those cytokines or chemokines” | --a cytokine or chemokine as-- |
| Column 39, line 21: | Page 51, line 10 (original claim 13, renumbered as claim 11): |
| “lipopolysaceharide” | --lipopolysaccharide--. |

A true and correct copy of pages 5, 15, 31, 33, 34, 38, and 51 of the specification as filed and a true and correct copy of Applicants' Amendment under 37 C.R.F. §1.111 dated January 29, 2007 which support Applicants' assertion of the errors on the part of the Patent Office accompanies this Certificate of Correction.

Approval of the Certificate of Correction is respectfully requested.

Respectfully submitted,

Frank C Eisenschenk

Frank C. Eisenschenk, Ph.D.
Patent Attorney
Registration No. 45,332
Phone No.: 352-375-8100
Fax No.: 352-372-5800
Address: P.O. Box 142950
Gainesville, FL 32614-2950

FCE/jb/sl

Attachments: Copy of pages 5, 15, 31, 33, 34, 38, and 51 of the specification
Copy of Amendment under 37 C.R.F. §1.111 dated January 29, 2007

**[0021]** In various embodiments, the subject invention provides microbial ARCs that are transformed with vectors comprising at least one heterologous gene encoding IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-15, Il-16, Il-18, IL-23, IL-24, erythropoietin, G-CSF, M-CSF, platelet derived growth factor (PDGF), MSF, FLT-3 ligand, EGF, fibroblast growth factor (FGF; *e.g.*, aFGF (FGF-1), bFGF (FGF-2), FGF-3, FGF-4, FGF-5, FGF-6, or FGF-7), insulin-like growth factors (*e.g.*, IGF-1, IGF-2); vascular endothelial growth factor (VEGF); interferons (*e.g.*, IFN-γ, IFN-α, IFN-β); leukemia inhibitory factor (LIF); ciliary neurotrophic factor (CNTF); oncostatin M; stem cell factor (SCF); transforming growth factors (*e.g.*, TGF-α, TGF-β1, TGF-β1, TGF-β1), or chemokines (such as, but not limited to, BCA-1/BLC-1, BRAK/Kec, CXCL16, CXCR3, ENA-78/LIX, Eotaxin-1, Eotaxin-2/MPIF-2, Exodus-2/SLC, Fractalkine/Neurotactin, GROalpha/MGSA, HCC-1, I-TAC, Lymphotactin/ATAC/SCM, MCP-1/MCAF, MCP-3, MCP-4, MDC/STCP-1, ABCD-1, MIP-1α, MIP-1β, MIP-2α/GROβ, MIP-3α/Exodus/LARC, MIP-3β/Exodus-3/ELC, MIP-4/PARC/DC-CK1, PF-4, RANTES, SDF1α, TARC, or TECK) or those cytokines and/or chemokines provided in Tables 1, 8 and 9. In a preferred embodiment, ARCs contain IFN-γ (*e.g.*, bovine, avian (*e.g.*, chicken), fish, or human IFN-γ). In another preferred embodiment, ARCs comprise IFN-γ and IFN-α (*e.g.*, bovine, avian (*e.g.*, chicken), fish, or human fish IFN-γ and IFN-α). As used herein, the terms "ARC" or "ARCs" indicate amended recombinant cells that contain one or more heterologous genes. Amended recombinant cells containing no heterologous interferon genes or interferon protein are referred to as "Control ARCs" or "ARC controls".

**[0022]** In some embodiments microbial cells co-express one or more other heterologous genes, encoding antigens and/or antigenic proteins. Non-limiting examples of antigens or antigenic proteins include, and are not limited to, autoantigens, tumor antigens, MMR vaccines, polio vaccines, tetanus vaccines, pathogens normally encountered by an individual in the environment (*e.g.*, food borne pathogens such as *Klebsiella*, *Salmonella*, *Escherichia* spp., hepatitis viruses, influenza viruses, etc.) and pathogenic substances specifically introduced into the environment of the individual, such as a biotoxin (*e.g.*, mycotoxins, such as trichothecene mycotoxin (T-2), Staphylococcal enterotoxin B, ricin, or *Clostridium botulinum* neurotoxin, weaponized microbial cells (*e.g.*, viruses containing toxin DNA or RNA inserts, or bacterial or fungal cells transformed with toxins [*e.g.*, mycotoxins, such as trichothecene mycotoxin (T-2), Staphylococcal enterotoxin B, ricin, or *Clostridium botulinum* neurotoxin], viral pathogens, fungal pathogens, or bacterial pathogens (*e.g.*, smallpox, anthrax, Ebola virus, *Yersinia pestis*), or administered in amounts effective to reduce the severity of disease or disease symptoms and/or prevent the onset of disease or disease symptoms. In certain embodiments, the ARCs contain IFN-γ.

**[0047]** Thus, the subject invention provides a number of non-limiting embodiments and aspects that include:

A) An amended recombinant cell (ARC) comprising one or more heterologous genes encoding a chemokines and/or a cytokine;

B) The ARC according to embodiment A, wherein the heterologous gene(s) encode(s) IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-15, Il-16, Il-18, IL-23, IL-24, erythropoietin, G-CSF, M-CSF, platelet derived growth factor (PDGF), MSF, FLT-3 ligand, EGF, fibroblast growth factor (FGF; *e.g.*, aFGF (FGF-1), bFGF (FGF-2), FGF-3, FGF-4, FGF-5, FGF-6, or FGF-7), insulin-like growth factors (*e.g.*, IGF-1, IGF-2); vascular endothelial growth factor (VEGF); interferons (*e.g.*, IFN-γ, IFN-α, IFN-β); leukemia inhibitory factor (LIF); ciliary neurotrophic factor (CNTF); oncostatin M; stem cell factor (SCF); transforming growth factors (*e.g.*, TGF-α, TGF-β1, TGF-β1, TGF-β1), or chemokines (such as, but not limited to, BCA-1/BLC-1, BRAK/Kec, CXCL16, CXCR3, ENA-78/LIX, Eotaxin-1, Eotaxin-2/MPIF-2, Exodus-2/SLC, Fractalkine/Neurotactin, GROalpha/MGSA, HCC-1, I-TAC, Lymphotactin/ATAC/SCM, MCP-1/MCAF, MCP-3, MCP-4, MDC/STCP-1, ABCD-1, MIP-1α, MIP-1β, MIP-2α/GROβ, MIP-3α/Exodus/LARC, MIP-3β/Exodus-3/ELC, MIP-4/PARC/DC-CK1, PF-4, RANTES, SDF1α, TARC, or TECK) or those cytokines and/or chemokines provided in Tables 1, 8 and 9;

C) The ARC according to any previous embodiment, wherein the heterologous gene encodes IFN-γ (*e.g.*, bovine, avian (*e.g.*, chicken), fish, or human IFN-γ);

D) The ARC according to any previous embodiment, wherein the ARC further comprises a heterologous gene encoding IFN-α (*e.g.*, bovine, avian (*e.g.*, chicken), fish, or human IFN-α);

E) The ARC according to any previous embodiment, further comprising one or more heterologous genes encoding autoantigens, tumor antigens, MMR vaccines, polio vaccines, tetanus vaccines, antigens associated with pathogens normally encountered by an individual in the environment (*e.g.*, food borne pathogens such as *Klebsiella*, *Salmonella*, *Escherichia* spp., hepatitis viruses, influenza viruses, etc.), pathogenic substances or antigens that may be

Each group consisted of six, 6-8 month old, Angus-Hereford-cross calves that were either females or neutered males. Data presented are the mean ± SEM and are illustrated in Figure 11.

**[0084]** Data for the secondary immune response are consistent with data for the primary immunization. The lowest dose of BGI/ARC (2.5 μg BGI) gave the greatest enhancement of antibody titers, whereas the ARC-minus, BGI control enhanced the immune response little, if at all. The difference between the control and the maximum titer increased from 27 fold in the primary response to over 150 fold in the secondary response. As illustrated in Figure 12, BGI/ARCs have a proliferative effect on lymphocytes (as measured by the incorporation of $^3$H thymidine).

**[0085]** The present examples illustrate that ARCs are valuable tools for the inexpensive production, preparation, and delivery of stabilized IFN-γ. IFN-γ/ARCs are also surprisingly useful as immunoadjuvants and accelerants of the immune response. As described herein: 1.) The *Pseudomonas fluorescens* expression system can be used to inexpensively produce prodigious amounts of active IFN-γ having an expression level equal to that obtained for a commercial insecticide, MVP® (9), protein (the process described herein can produce more than one ton of IFN-γ from a single 100,000-liter fermentation); 2.) The chemical sterilization procedure amends the *Pseudomonas* cells, stabilizes the IFN-γ contents of ARCs, and provides for the effective release of IFN-γ within or on the surface of a macrophage or other IFN-γ reactive cell; 3.) The amended and stabilized BGI/ARCs are active in their intact form, and picogram levels of IFN-γ protect cells from infection by virus (see, for example, VSV infection of bovine kidney cells); 4.) Unlike many other recombinant proteins, IFN-γ is soluble and can be over-expressed in *P. fluorescens* in soluble form and does not form inclusion bodies in cells even when expressed at levels greater than 40% of total cell protein; 5.) BGI/ARCs have excellent shelf-life properties, remaining stable and active after several weeks at 37°C and remain active for more than 6 months without loss of activity when frozen; 6.) BGI/ARCs have exceptional physical properties; they are mechanically durable, nonflocculant microscopic particles that can remain in suspension and easily pass through a syringe; 7.) BGI/ARCs have desirable time-release properties; 8) Microgram quantities of IFN-γ in BGI/ARCs produce an unexpectedly strong response in cattle and promote unexpectedly vigorous immunoadjuvant responses and immunoacceleration, and 9.) BGI/ARCs can be given intramuscularly, subcutaneously, or introduced to the body through mucosal membranes, making non-invasive delivery possible. The

Example 12 – Avian administration of ARCs containing chicken IFN-γ

**[0089]** CGI/ARCs were prepared as described above and HN plant-derived antigen was prepared according to methods known in the art (see, for example, U.S. Patent No. 5,310,678 and U.S. Provisional Application 60/467,998, filed May 5, 2003, which are hereby incorporated by reference in its entirety) with the following modification. Plant derived antigen NT1 cells were harvested 6-12 days after passage. Whole wet NT1 cells harvested directly from cell culture were filtered to remove excess media by placing a Spectramesh 30 filter in a Buchner funnel and pouring cells and media through the filter using a slight vacuum.

**[0090]** To make a preparation of HN vaccine material for assay detection, 0.5 grams of cells were placed in 2 mL of extraction buffer (Dulbecco's phosphate buffered saline (DPBS), 1mM EDTA, pH 7.2)), and then sonicated for about 2 minutes on ice. Sonication was performed using a Branson 450 sonifier with a replaceable microtip at output control of 8, duty cycle 60 for 2 minutes (for larger preparations, (>5 grams) sonication was performed for 5 -10 minutes on ice). Sonicates were then placed on ice until use. Inactivated NDV La Sota strain was derived from allantoic fluid (Lohman Animal Health) at a pre-inactivation egg titer $\geq 10^{10.6}$ $EID_{50}$/mL. The allantoic fluid was stored as a frozen preparation (-80°C) until use.

**[0091]** For vaccination, SPF chicks from SPAFAS (North Franklin, Conn.) were obtained at one-day of age and placed in cages and allowed to acclimate until 7 days of age. The number of chicks per treatment was based on a completely randomized design using repeat measurements. Any excess chicks were placed randomly in individual cages and were utilized to replace chicks that died from shipping or placement stress. Subcutaneous inoculation was performed by injecting 0.1-0.25 mL into nape of the neck.

**[0092]** Dose and administration of antigen and CGI/ARCs was performed as follows. Plant-derived samples were prepared by hydrating freeze dried CHN extracts in DPBS with 25 μg of CGI/ARC material. Inactivated allantoic fluid was thawed and mixed by adding 25μg of CGI/ARC directly to the sample. For plant-derived samples containing oil/water emulsion samples, the freeze dried material was resuspended directly into in DPBS containing 0.5% Tween and 2.5% Drakeol Oil with 0.165% Span 80. Two inoculations of antigen were administered, (day 0 at 7-days of age) and a second booster dose at day 14 (21-days of age), the birds were then given an inactivated NDV-infected allantoic fluid (described above) at day 35 (42 days of age) and the trial was terminated at day 42 (49 days of age). A 1 to 2 mL blood sample was collected from each bird via venipuncture of either the jugular or peripheral wing vein on days 14, 21, 35 and 42 of the study.

**[0093]** To measure the immune response, chicken red blood cells in Alsever's solution (CRBC) were obtained from Colorado Serum (L#8152). To prepare a 1% solution of CRBCs, five mL was transferred to a 15 mL conical tube and centrifuged at 250 x g for 10 minutes. The supernatant and buffy coat were pipetted from the RBC pellet; the pellet was washed twice by resuspending in 1 x DPBS (Dulbecco's Phosphate Buffered Saline) (L# 003435E JRH) and centrifuged 250 x g for 10 minutes. The pellet was resuspended to 1% (v/v) in DPBS. To confirm the concentration of the suspension, 400µl was transferred to 1.6 mL of deionized water and cells lysed by mixing vigorously. The $OD_{540}$ was between 0.4 – 0.5. The 1% solutions were stored at 2-7°C until used.

**[0094]** To test hemagglutination, a 96-well U-bottom dish (Falcon) was first sprayed with Static Guard® and blotted onto paper towels. Virus samples were prediluted in DPBS 1:2 and 50 µl of DPBS were placed to each well of the 96-well dish. The diluted virus was added to the first row and then serially diluted 2-fold for the desired number of dilutions per virus sample. 50µl of 1% CRBC was added to each well and the plate was mixed for 20 seconds at 600 rpm. The plate was placed on wet paper towels and incubated until the CRBCs in the control wells (DPBS and CRBCs at 1:1 ratio) pellet to the bottom of the plate, or for at least 1 hr at 2-7°C. The end point was the dilution of the last well in the series that provides 100% agglutination.

**[0095]** Virus was prediluted in DPBS to provide 4-8 HA units per 50 µl (based on titering the virus described above). A separate plate was set up using 25 µl of DPBS per well in columns 1 and 3-12; 25 µl of serum was added per well in column 1 and 3; serum in column 3 was serially diluted 2 fold through 10 wells. The pretitered virus (25 µl) was then added in all wells column 3-12 and mixed 20 seconds at 600rpm; the plate was allowed to incubate at room temperature for 1h +/- 15 minutes. Fifty µl of 1% CRBC was then added per well, mixed 20 seconds at 600 rpm and incubated in a humidifying chamber overnight at 2-7°C for AIV or 1-2 hours at 2-7°C for NDV. The titer of the serum is the last well in the series dilution that inhibits agglutination 100%.

**[0096]** The serum geometric mean titer (GMT) was determined for each treatment group using Microsoft Excel 2000 version 9.0.3821 SR-1. Background ELISA titers of < 10 were given

26. **Zuffa, A. and N. Feketeova**. 1980. Protection of cattle vaccinated with inactivated oil adjuvant infectious bovine rhino tracheitis vaccine against experimental infection. **27**:725-733.

11. A method of inducing or accelerating an immune response in an individual to an antigen or immunogen comprising the step of administering, to an individual:

amended recombinant cells (ARCs) comprising at least one heterologous gene encoding a chemokine or a cytokine; or

a composition comprising amended recombinant cells (ARCs) comprising at least one heterologous gene encoding a chemokine or a cytokine.

12. The method according to claim 11, wherein said method further comprises the administration of an antigen of interest.

13. The method according to claim 12, further comprising the administration of lipopolysaccharide (LPS).

14. The method according to claim 11, wherein said heterologous gene encodes IFN-γ.

15. The method according to claim 14, wherein said IFN-γ is bovine, avian, fish, or human.

16. The method according to claim 15, wherein said IFN-γ is bovine.

17. The method according to claim 15, wherein said avian IFN-γ is chicken IFN-γ.

18. The method according to claim 11, wherein the ARCs co-express at least one antigen of interest.

19. A method of accelerating the immune response of an individual to an antigen or immunogen comprising the administration of:

a) amended recombinant cells (ARCs) comprising at least one heterologous gene encoding a chemokine or a cytokine; or

b) a composition comprising amended recombinant cells (ARCs) comprising at least one heterologous gene encoding a chemokine or a cytokine;

to an individual in amounts effective to accelerate the immune response of the individual.

20. The method according to claim 19, wherein said method accelerates the development of IgM, IgG, IgA, IgE, or IgY antibodies.

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Frank C. Eisenschenk, Ph.D., Patent Attorney

AMENDMENT UNDER 37 C.F.R. § 1.111
Examining Group 1636
Patent Application
Docket No. DAS-103XC1
Serial No. 10/681,540

## IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Examiner : Nancy S. Vogel
Art Unit : 1636
Applicants : Frank H. Gaertner, Stacey Finstad Lee, Robert Shutter
Serial No. : 10/681,540
Filed : October 7, 2003
Conf. No. : 7752
For : Amended Recombinant Cells for the Production and Delivery of an Antiviral Agent, Adjuvant and Vaccine Accelerant

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313

## AMENDMENT UNDER 37 C.F.R. § 1.111

Sir:

A Petition and Fee for a one-month Extension of Time through and including January 29, 2007, accompanies this Amendment.

In response to the Office Action dated September 29, 2006, please amend the above-identified patent application as follows:

## In the Specification

Please substitute paragraph [0005] beginning on page 2 as follows:

In one aspect of the invention, IFN-γ, as well as other cytokines, can be expressed in a wide variety of microbial cells, including the bacterium *Pseudomonas fluorescens*, through the use of genetic engineering techniques well-~~know~~ known in the art. Appropriately reconstructing the cytokine gene and positioning it precisely in a host plasmid-vector between a strong regulated promoter and transcription/translation terminators, routinely accomplishes the expression of IFN-γ in a particular foreign host. The suitability of any such host is also routine to test by ordinary means by one of ordinary skill in the art, without undue experimentation. Once the transformed microbial cells have expressed the IFN-γ or other cytokines to a high level, the cells can be amended with agents, including heat and chemical agents, that kill (sterilize) the cells, stabilize an expressed active cytokine, and amend the cell wall for optimal release of the cell-encased cytokine. For example, the sterilization/amendment procedure used in the present description for the cells of *Pseudomonas fluorescens*, expressing IFN-γ, is a modification of a process originally used for the production of a commercial biopesticide, MVP (4;9;19). According to this method, the cells of *P. fluorescens* are treated with Lugol iodine at a pH of about 4.3. This pH thoroughly sterilizes the culture *in situ*, amends the cell wall for physical durability, and renders the bacterial cell wall susceptible to proteolytic dissolution (9). The procedure also eliminates cellular flocculation, and appears to decrease the endotoxic properties of the *Pseudomonas* cell wall.

Please substitute paragraph [0017] beginning on page 4 as follows:

**Figure 11** illustrates the immunoadjuvant activity of BGI/ARCs and the ~~ability o~~ ability of BGI-ARCs to accelerate the immune response of calves to an antigen (*e.g.*, porcine serum albumin).

Please substitute the following paragraph on page 20, beginning at subsection HH):

HH) In various implementations of the methods of embodiments M through GG, compositions according to the subject invention can be administered orally, parenterally, as sprays (including inhalation sprays), topically, rectally, nasally, buccally, vaginally or via an implanted reservoir. The term parenteral, as used herein, includes subcutaneous, intradermal, intravenous, intrastriatial, intramuscular, intraperitoneal, intrathecal, intraventricular, ~~intrasternal ,or~~ intrasternal, or intracranial injection and other infusion techniques;

Please substitute Reference No. 24 beginning on page 37 as follows:

24. **Yilma, T., S. Owens, E. H. Fennie, and K. P. Anderson** . 1989. ~~Enahncement~~ Enhancement of primary and Secondary Immune Responses by Interferon-gamma. Adv.Exp.Med.Biol. **251**:145-152.

Please substitute the following Table 8 beginning on page 45 through to page 46:

## TABLE 8:

## CHEMOKINES

| **Reference** | **Protein** | **Function** |
|---|---|---|
| BCA-1 / BLC-1 | B Cell-Attracting chemokine-1 (B-Lymphocytes Chemoattracting) (CXCL13) | B-cell attractant |
| BRAK / Kec | CXC chemokine in breast and kidney / Kidney-expressed chemokine (SCYB14, CXCL14) | Involved in MΦ development |
| CXCL16 | CXC-chemokine 16 | |
| ENA-78 / LIX | Epithelial cell-derived neutrophil-activating protein 78 (CXCL5, SCYB5) | Neutrophil activating peptide |
| Eotaxin-1 | Eotaxin-1 (CCL11) | Eosinophil chemotaxis |
| Eotaxin-2 / MPIF-2 | Eotaxin-2 (CCL24,CKß6) | Chemotactic agent for T-cells and eosinophils |
| Exodus-2 / SLC | Exodus-2 (CCL21, CKß9, SCYA21) | Angiostatic activity, chemotacitc agent for T-cells, dendritic cells, CD34$^+$ hematopoietic cells, NK cells, and B-cells |
| Fractalkine/Neurotactin | Fractalkine / Neurotactin (CX3CL1) | Chemotactic agent for T-cells and monocytes |
| GROalpha / MGSA | Melanoma Growth Stimulatory Activity protein (CXCL1) | Neutrophil activation |
| HCC-1 | Hemofiltrate CC chemokine 1 (SCYA14, CCL14) | Chemotactic agent for monocytes and THP-1 cells |
| IL8 | Interleukin 8 (CXCL8) | Chemoattraactant for neutrophils, basophils, and T-cells; activates neutrophils |
| I-TAC | Interferon-stimulated T-cell alpha chemoattractant (CXCL11) | |
| Lymphotactin / ATAC / SCM | Lymphotactin (CL1, LTN) | Chemoattractant for T and NK cells |

| Reference | Protein | Function |
|---|---|---|
| MCP-1 / MCAF | Monocyte Chemotactic Protein 1 (CCL2, SCYA2) | Chemoattractant for monocytes and neutrophils; ~~aguments~~ augments neutrophil anti-tumor activity |
| MCP-3 | Monocyte Chemotactic Protein 3 (CCL7, SCYA7) | Chemoattractant for monocytres and eosinophils; augments monocyte anti-tumor activity |
| MCP-4 | Monocyte Chemotactic Protein 4 (CCL13, SCYA13) | Chemoattractant for monocytes, lymphocytes, basophils, and eosinophils |
| MDC / STCP-1 / ABCD-1 | Macrophage-Derived Chemokine (CCL22, SCYA22) | Chemoattractant for T-cells, activated lymphocytes, and monocytes |
| MIP-1α | Macrophage Inflammatory Protein 1 alpha (CCL3, SCYA3) | Chemoattractant for lymphocytes |
| MIP-1β | Macrophage Inflammatory Protein 1 beta (CCL4, SCYA4) | Chemoattractant for ~~moneytes~~ monocytes, ~~densritic~~ dendritic cells, NK cells and T-cells |
| MIP-2α / GROß | Macrophage Inflammatory Protein 2 alpha (CXCL2) | |
| MIP-3α / Exodus / LARC | Macrophage Inflammatory Protein 3 alpha (CCL20, SCYA20) | Chemoattractant for lymphocytes, activated NK cells, dendritic cells |
| MIP-3β / Exodus-3 / ELC | Macrophage Inflammatory Protein 3 beta (CCL19, SCYA19) | Chemoattractant for T-cells, B-cells, and dendritic cells |
| MIP-4 / PARC / DC-CK1 | Macrophage Inflammatory Protein 4 (CCL18, CKß7, SCYB18) | Chemoattractant for T-cells |
| RANTES | Rantes, formerly "T cell-specific protein" (CCL5) | Chemoattractant for memory T-cells, monocytes, and eosinophils |
| SDF1α | Stroma Cell-Derived Factor 1 Alpha | Chemoattractant for neutrophils, lymphocytes, and monocytes |
| TARC | Thymus and Activation-Regulated Chemokine (CCL17) | Chemoattractant for activated $T_{H2}$-cells |
| TECK | Thymus-Expressed Chemokine (CCL25) | Chemoattractant for thymocytes, MΦ, Thp-1 cells, and dendritic cells |

Please substitute the following Table 9 beginning on page 47:

## TABLE 9:

## CYTOKINES

| **Reference** | **Protein** | **Function** |
|---|---|---|
| GM-CSF | Macrophage Colony Stimulating Factor | Growth and differentiation of hematopoietic lineages (*e.g.*, granulocytes, MΦ, eosinophils, and erythrocytes |
| IFN alpha | Interferon Alpha | Anti-tumor and anti-viral activity |
| IFN beta | Interferon Beta | Antiviral, antibacterial, and anticancer activity |
| IFN gamma | Interferon Gamma | Stimulates CTL responses; antiviral and anti-proliferative activity on transformed cells |
| **Interleukins** | | |
| IL-1 beta | Interleukin-1 Beta | Stimulates B-cell maturation/proliferation |
| IL-2 | Interleukin-2 | Regulates immune response and T-cell proliferation |
| IL-4 | Interleukin-4 | Activates B-cells |
| IL-6 | Interleukin-6 | B-cell differentiation |
| IL-10 | Interleukin-10 | ~~Immunosupressive~~ immunosuppressive and anti-inflammatory |
| IL-12elasti | Interleukin-12 (with elastin linker between subunits) | Growth factor for activated T-cells and MNK cells; enhances lytic activity of NK/LAK cells |
| IL-13 | Interleukin-13 | Anti-inflammatory agent |
| IL-15 | Interleukin-15 | Stimulates T-lymphocyte and NK proliferation |
| IL-16 | Interleukin-16 | Chemoattractant for CD4 lymphocytes, monocytes, dendritic cells, and eosinophils |
| IL-18 | Interleukin-18 | Induces IFN-γ and augments NK activity |
| IL-18BPa | Interleukin-18 binding protein, isoform A | Inhibitor of early Th1 cytokine response |
| IL-23 | Interleukin-23 | Stimulates proliferation of memory T-cells; stimulates IFN-γ production |
| IL-24 | Interleukin-24 | |
| VIP | Vasoactive Intestinal Peptide | Vasodialator; lowers arterial blood pressure; stimulates myocardial contractility; smooth muscle relaxant; MΦ activator; stimulates T-cell proliferation |
| **TNF Superfamily** | | |
| LIGHT / TNFSF14 | Tumor Necrosis Factor SuperFamily member 14 | Induces apoptosis, stimulates T-cells, suppresses *in vivo* tumor formation |
| sTALL-1 / | Tumor Necrosis Factor | Stimulates B-cell proliferation |

| | | |
|---|---|---|
| TNFSF13B<br>(also called BLyS, BAFF, THANK) | SuperFamily member 13B | |
| TNFalpha / TNFSF2 | Tumor Necrosis Factor Alpha | Cytolysis of tumor cells; induces cell differentiation |
| TWEAK / TNFSF12 | Tumor Necrosis Factor SuperFamily member 12 (also called Apo3L) | Inces tumor cell death, influences astrocyte behavior |

## In the Claims

Claim 1 (Currently amended).

An amended recombinant cell (ARC) comprising at least one heterologous gene, said at least one heterologous gene encoding a chemokine or a cytokine.

Claim 2 (Currently amended).

The ARC according to claim 1, wherein said ARC further comprises at least one additional ~~the~~ heterologous gene that encodes IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-15, Il-16, Il-18, IL-23, IL-24, erythropoietin, G-CSF, M-CSF, platelet derived growth factor (PDGF), MSF, FLT-3 ligand, EGF, fibroblast growth factor (FGF); aFGF (FGF-1); bFGF (FGF-2); FGF-3; FGF-4; FGF-5; FGF-6; FGF-7; insulin-like growth factor 1 (IGF-1); IGF-2; vascular endothelial growth factor (VEGF); IFN-γ; IFN-α; IFN-β; leukemia inhibitory factor (LIF); ciliary neurotrophic factor (CNTF); oncostatin M; stem cell factor (SCF); TGF-α; TGF-β1; TGF-β2; a chemokine selected from the group consisting of BCA-1/BLC-1, BRAK/Kec, CXCL16, CXCR3, ENA-78/LIX, Eotaxin-1, Eotaxin-2/MPIF-2, Exodus-2/SLC, Fractalkine/Neurotactin, GROalpha/MGSA, HCC-1, I-TAC, Lymphotactin/ATAC/SCM, MCP-1/MCAF, MCP-3, MCP-4, MDC/STCP-1, ABCD-1, MIP-1α, MIP-1β, MIP-2α/GROβ, MIP-3α/Exodus/LARC, MIP-3β/Exodus-3/ELC, MIP-4/PARC/DC-CK1, PF-4, RANTES, SDF1α, TARC, and TECK; or ~~those cytokines or chemokines~~ a cytokine or chemokine as provided in Tables 1, 8, and 9.

Claim 3 (Currently amended).

The ARC according to ~~claim 2~~claim 1, ~~wherein the~~ wherein said at least one heterologous gene encodes IFN-γ.

Claim 4 (Original).

The ARC according to claim 3, wherein said IFN-γ is bovine, avian, fish, or human.

Claim 5 (Original).

The ARC according to claim 4, wherein said IFN-γ is bovine.

Claim 6 (Original).

The ARC according to claim 4, wherein said avian IFN-γ is chicken IFN-γ.

Claim 7 (Currently amended).

The ARC according to claim 2, wherein ~~the ARC further comprises a heterologous gene encoding~~ said at least one additional heterologous gene encodes IFN-α.

Claim 8 (Original).

The ARC according claim 1, wherein said cell is a microbial cell selected from the group consisting of Gram positive organisms, Gram negative organisms, yeast, and fungi.

Claim 9 (Original).

The ARC according to claim 8, wherein the microbial cell is *Pseudomonas fluorescens*.

Claim 10 (Original).

The ARC according to claim 1, further comprising a carrier.

Claim 11 (Withdrawn).

A method of inducing or accelerating an immune response in an individual to an antigen or immunogen comprising the step of administering, to an individual:

amended recombinant cells (ARCs) comprising at least one heterologous gene encoding a chemokine or a cytokine; or

a composition comprising amended recombinant cells (ARCs) comprising at least one heterologous gene encoding a chemokine or a cytokine.

Claim 12 (Withdrawn).

The method according to claim 11, wherein said method further comprises the administration of an antigen of interest.

Claim 13 (Withdrawn).

The method according to claim 12, further comprising the administration of lipopolysaccharide (LPS).

Claim 14 (Withdrawn).

The method according to claim 11, wherein said heterologous gene encodes IFN-γ.

Claim 15 (Withdrawn).

The method according to claim 14, wherein said IFN-γ is bovine, avian, fish, or human.

Claim 16 (Withdrawn).

The method according to claim 15, wherein said IFN-γ is bovine.

Claim 17 (Withdrawn).

The method according to claim 15, wherein said avian IFN-γ is chicken IFN-γ.

Claim 18 (Withdrawn).

The method according to claim 11, wherein the ARCs co-express at least one antigen of interest.

Claim 19-31 (Canceled).

Claim 32 (Currently amended).

A method of making an amended recombinant cell (ARC) comprising the steps of:

a) introducing at least one heterologous gene encoding a cytokine into a cell ~~and, optionally, a chemokine into a cell~~;

b) growing said cell in a nutrient medium;

c) harvesting said cells; and

d) inactivating or fixing said cells.

Claim 33 (Currently amended).

The method according to claim 32, said method comprising introducing at least one additional ~~wherein the~~ heterologous gene into said cell, said at least one additional heterologous gene ~~encodes~~ encoding IL-1, IL-2, IL-3, IL-4, IL-5, IL-6, IL-7, IL-8, IL-9, IL-10, IL-11, IL-15, Il-16, Il-18, IL-23, IL-24, erythropoietin, G-CSF, M-CSF, platelet derived growth factor (PDGF), MSF, FLT-3 ligand, EGF, fibroblast growth factor (FGF); aFGF (FGF-1); bFGF (FGF-2); FGF-3; FGF-4; FGF-5; FGF-6; FGF-7; insulin-like growth factor 1 (IGF-1); IGF-2; vascular endothelial growth factor (VEGF); IFN-γ;-IFN-α; IFN-β; leukemia inhibitory factor (LIF); ciliary neurotrophic factor (CNTF); oncostatin M; stem cell factor (SCF); TGF-α; TGF-β1; TGF-β2; a chemokine selected from the group consisting of BCA-1/BLC-1, BRAK/Kec, CXCL16, CXCR3, ENA-78/LIX, Eotaxin-1, Eotaxin-2/MPIF-2, Exodus-2/SLC, Fractalkine/Neurotactin, GROalpha/MGSA, HCC-1, I-TAC, Lymphotactin/ATAC/SCM, MCP-1/MCAF, MCP-3, MCP-4, MDC/STCP-1, ABCD-1, MIP-1α, MIP-1β, MIP-2α/GROβ, MIP-3α/Exodus/LARC, MIP-3β/Exodus-3/ELC, MIP-4/PARC/DC-CK1, PF-4, RANTES, SDF1α, TARC, and TECK; or ~~those cytokines or chemokines~~ a cytokine or chemokine as provided in Tables 1, 8 and 9.

Claim 34 (Currently amended).

The method according to ~~claim 33~~claim 32, wherein ~~the heterologous~~ said at least one heterologous gene encodes IFN-γ.

Claim 35 (Original).

The method according to claim 34, wherein said IFN-γ is bovine, avian, fish, or human.

Claim 36 (Original).

The method according to claim 35, wherein said IFN-γ is bovine.

Claim 37 (Original).

The method according to claim 35, wherein said avian IFN-γ is chicken IFN-γ.

Claim 38 (Currently amended).

The method according to claim 33, wherein ~~the ARC further comprises a heterologous gene encoding~~ said at least one additional heterologous gene encodes IFN-α.

Claim 39 (New).

The method according to claim 34, said method comprising introducing at least one additional heterologous gene encoding a chemokine into said cell.

Claim 40 (New).

The method according to claim 34, wherein said at least one additional heterologous gene encodes IFN-α.

Claim 41 (New).

The ARC according to claim 3, wherein said ARC further comprises at least one additional heterologous gene that encodes IFN-α.

Claim 42 (New).

The ARC according to claim 9, wherein said *Pseudomonas fluorescens* cell comprises at least one heterologous gene that encodes IFN-γ.

Claim 43 (New).

The ARC according to claim 42, wherein said IFN-γ is bovine, avian, fish, or human.

Claim 44 (New).

The ARC according to claim 43, wherein said IFN-γ is bovine.

Claim 45 (New).

The ARC according to claim 43, wherein said avian IFN-γ is chicken IFN-γ.

Claim 46 (New).

The ARC according to claim 42, wherein said ARC further comprises at least one additional heterologous gene.

Claim 47 (New).

The ARC according to claim 46, wherein said at least one additional heterologous gene. encodes IFN-α.

## Remarks

Claims 1-38 are pending in the subject application. By this Amendment, Applicants have canceled claims 19-31, amended claims 1-3, 7, 32, 33, 34, and 38 and added new claims 39-47. Applicants have also amended the subject specification to correct inadvertent typographical errors. Support for the amendments and new claims can be found throughout the subject specification and in the claims as originally filed (see, for example, paragraph 47 of the as-filed application). Entry and consideration of the amendments presented herein is respectfully requested. Accordingly, claims 1-33 and 35-46 are currently before the Examiner. Favorable consideration of the pending claims is respectfully requested.

Claims 2 and 33 have been objected to on the basis of various informalities. The Office Action indicates that these claims have an inconsistency with respect to the recitation of "or those cytokines or chemokines provided in Tables 1, 8 or 9" as this phrase is inconsistent with the singular form of the claim structure as relates to the recitation of "the heterologous gene encodes". Applicants thank the Examiner for her careful review of the claims and have amended the claims to attend to this issue in the manner suggested in the Office Action. Accordingly, reconsideration and withdrawal of this objection is respectfully requested.

Claims 7 and 38 have been objected to under 37 C.F.R. §1.75(c) as being of improper dependent form for failing to further limit the subject matter of a previous claim. Applicants have amended the claims in an effort to address this issue and now respectfully submit that these claims further limit the subject matter of the claims from which they depend. As the Examiner will note, the claims have been amended to indicate that the claims now indicate that the method or ARC further comprises IFN-α as a second heterologous gene or gene product. Thus, claims 7 and 38 now further limit the claims from which they, respectively, depend. Accordingly, reconsideration and withdrawal of the rejection is respectfully requested.

Claims 1 and 2 have been rejected under 35 U.S.C. §102(b) as being anticipated by Tarnowski. The Office Action argues that the cells disclosed meet the definition for an ARC since acid treatment is disclosed in the subject specification as one means for the production of the ARCs.

Applicants respectfully submit that the cells of Tarnowski do not meet the definition of an ARC. Applicants respectfully submit that the cells of Tarnowski appear to be easily disrupted by passage through a homogenizer at about 6000 psi and about 1000 psi (see column 12, Example 4) and the cells produce an interferon that is present in an oligomeric form. Applicants also respectfully submit that Tarnowski fails to anticipate the claimed invention as it is directed to purifying oligomeric forms of recombinant interferons from bacterial cells and states that recombinant interferons must be purified, preferably to homogeneity, before being employed as a therapeutic agent (see column 1, lines 28-34). Tarnowski further states that the purification methods favor the formation of interferon oligomers (column 1, lines 34-42) and that the oligomeric forms have, in many cases, no biological activity, lower activity, or cause deleterious side effects when administered therapeutically (column 1, lines 44-46). In contrast, ARCs of the instant invention have enhanced physical durability and resist disruption by sonic oscillation or rupture through a French Pressure Cell (see paragraph 30, page 8 of the as-filed specification). Additionally, the ARCs induce at least one desired biological effect in an individual (see paragraph 42, page 13 of the as-filed specification). Finally, Applicants respectfully submit that the cells of Tarnowski would not exhibit the stability associated with ARCs of the subject invention as illustrated in Figure 5. Accordingly, it is respectfully submitted that Tarnowski does not anticipate the claimed invention as the cells taught therein cannot be used in the same manner as those of the instant invention. Thus, reconsideration and withdrawal of the rejection is respectfully requested.

Claims 1, 2, 8, 10, 32 and 33 have been rejected as being anticipated by Steidler *et al.* (*Infect. Immun.*, 1998, 66(7):3183-3189). The Office Action argues that Steidler *et al.* teach recombinant cells (*L. lactis*) comprising at least one heterologous gene encoding a cytokine (IL-2 or IL-6) and further disclose that the cells have been killed with mitomycin C. The Office Action further argues that the reference is considered to meet the definition of an ARC since the reference teaches the treatment of *L. lactis* with an antibiotic (mitomycin C) and antibiotic treatment is one of the means taught in the specification for producing ARCs. Applicants traverse and respectfully submit that the cells of Steidler et al. do not meet the definition of an ARC.

As noted in the specification, the subject invention provides **active** (emphasis added) cytokine and/or chemokine compositions that have been expressed in processed (amended) microbial systems (ARCs). ARCs are recombinant microbial cells containing expressed, heterologous proteins, the cells of which have been killed through specific chemical-sterilization processes that amend the cell wall of the microbial cells. Further, ARCs maintain the desired biological activity of the heterologous gene contained therein (see paragraph 20, page 4 and paragraph 30, page 8).

Steidler *et al.* evaluated whether the antibody response *in vivo* is dependent on the production of active cytokine rather than being associated with the release of preformed bacterially associated cytokine in the animal. The reference teaches that treatment of the bacterial cells with mitomycin C abrogated the immunostimulatory effect of the IL-2 or IL-6 and that live cytokine secreting strains of bacteria were required to cause the desired biological effects (see Abstract, lines 8-10, page 2187; "Influence of lactococcal cell viability on immune responses elicited by recombinant *L. lactis*", page 3187, column 1; and Figure 6). Thus, it is respectfully submitted that the inactivated cells of Steidler *et al.* do not provide an active form of the cytokine when the cells were treated with the antibiotic and the reference does not anticipate the claimed invention. Accordingly, reconsideration and withdrawal of the rejection is respectfully requested.

Claims 1-10 and 32-38 are rejected under 35 U.S.C. §112, first paragraph, as failing to comply with the written description requirement. The Office Action argues that the disclosure is not deemed to be descriptive of the complete structure of a representative number of species encompassed by the claims as one of skill in the art cannot envision all the methods of producing the claimed ARCs based on the teachings of the specification. Namely, the Office Action argues that there is insufficient disclosure of types of treatment that would be useful for different types of cells and it is argued that the specification fails to describe the actual structures responsible for the properties which define the ARC cells. Therefore, it is argued, the specification does not describe the claimed ARC cells and the method of making the ARC cells in such full, clear, concise and exact terms so as to indicate that Applicants were in possession of the method at the time of filing of the present application. Applicants traverse.

Applicants respectfully submit that the as-filed specification provides adequate written description for the claimed invention. An objective standard for determining compliance with the written description requirement is, "does the description clearly allow persons of ordinary skill in the art to recognize that he or she invented what is claimed." *In re Gosteli*, 872 F.2d 1008, 1012, 10 USPQ2d 1614, 1618 (Fed. Cir. 1989). Under *Vas-Cath, Inc. v. Mahurkar*, 935 F.2d 1555, 1563-64, 19 USPQ2d 1111, 1117 (Fed. Cir. 1991), to satisfy the written description requirement, an applicant must convey with reasonable clarity to those skilled in the art that, as of the filing date sought, he or she was in possession of the invention, and that the invention, in that context, is whatever is now claimed. As indicated in *Capon v. Eshhar*, 418 F.3d 1349, 1357, 76 U.S.P.Q.2d 1078, 1084 (Fed. Cir. 2005),

> The "written description" requirement implements the principle that a patent must describe the technology that is sought to be patented; the requirement serves both to satisfy the inventor's obligation to disclose the technologic knowledge upon which the patent is based, and to demonstrate that the patentee was in possession of the invention that is claimed. *See Enzo Biochem*, 296 F.3d at 1330 (the written description requirement "is the quid pro quo of the patent system; the public must receive meaningful disclosure in exchange for being excluded from practicing the invention for a limited period of time"); *Reiffin v. Microsoft Corp.*, 214 F.3d 1342, 1345-46 (Fed. Cir. 2000) (the purpose of the written description requirement "is to ensure that the scope of the right to exclude . . . does not overreach the scope of the inventor's contribution to the field of art as described in the patent specification"); *In re Barker*, 559 F.2d 588, 592 n.4 (C.C.P.A. 1977) (the goal of the written description requirement is "to clearly convey the information that an applicant has invented the subject matter which is claimed"). The written description requirement thus satisfies the policy premises of the law, whereby the inventor's technical/scientific advance is added to the body of knowledge, as consideration for the grant of patent exclusivity.
>
> The descriptive text needed to meet these requirements varies with the nature and scope of the invention at issue, and with the scientific and technologic knowledge already in existence. The law must be applied to each invention that enters the patent process, for each patented advance is novel in relation to the state of the science. Since the law is applied to each invention in view of the state of relevant knowledge, its application will vary with differences in the state of knowledge in the field and differences in the predictability of the science.

Additionally, the Federal Circuit has indicated that "(1) examples are not necessary to support the adequacy of a written description; (2) the written description standard may be met (as it is here) even where actual reduction to practice of an invention is absent; and (3) there is no *per se* rule that an adequate written description of an invention that involves a biological macromolecule must contain a recitation of known structure" (see *Falko-Gunter Falkner v. Inglis*, 448 F.3d 1357, 1366 (Fed. Cir. 2006)). In the case of the instant invention, Applicants respectfully submit that the as-filed specification meets the written description requirements required under 35 U.S.C. § 112, first paragraph and that description clearly allows persons of ordinary skill in the art to recognize that the inventors were in possession of what is claimed.

As noted in the Office Action, the specification indicates that process for the manufacture of ARCs results in the alteration of cell wall properties by denaturing proteins in the cell wall and the strengthening of the cell walls (paragraph 20, page 4 of the as-filed application). Applicants respectfully submit that one skilled in the art, at the time the invention was made, would have been able to recognize that the inventors were in possession of what is claimed. Particularly, it is respectfully submitted that the scientific knowledge already in existence at the time the instant application was made would have indicated those agents that could confer the desired properties on cells within the scope of the instant claims. As indicated within McDonnell and Russell (*Clin. Microbiol. Rev.*, 1999, 12(1):147-179), a copy of which is attached for the Examiner's convenience, mechanisms of action of a number of materials identified in the as-filed specification have been examined and a battery of techniques are available for studying the mechanisms of action in microorganisms (see page 148, column 2). Additionally, compounds capable of causing such effects were also reported in the literature (*e.g.*, glutaraldehyde, formaldehyde, halogenating agents; see reference 9 as cited in this application and McDonnell and Russell, page 151-153). Accordingly, it is respectfully submitted that the as-filed specification provides adequate written description for the claimed invention and reconsideration and withdrawal of the rejection is respectfully requested.

It should be understood that the amendments presented herein have been made solely to expedite prosecution of the subject application to completion and should not be construed as an indication of Applicants' agreement with or acquiescence in the Examiner's position. Applicants expressly reserve the right to pursue the invention(s) disclosed in the subject application, including any subject matter canceled or not pursued during prosecution of the subject application, in a related application.

In view of the foregoing remarks and amendments to the claims, Applicants believe that the currently pending claims are in condition for allowance, and such action is respectfully requested.

The Commissioner is hereby authorized to charge any fees under 37 CFR §§1.16 or 1.17 as required by this paper to Deposit Account No. 19-0065.

Applicants invite the Examiner to call the undersigned if clarification is needed on any of this response, or if the Examiner believes a telephonic interview would expedite the prosecution of the subject application to completion.

Respectfully submitted,

Frank C. Eisenschenk, Ph.D.
Patent Attorney
Registration No. 45,332
Phone No.: 352-375-8100
Fax No.: 352-372-5800
Address: P.O. Box 142950
Gainesville, FL 32614-2950

FCE/sl

Attachment: McDonnell and Russell, *Clin. Microbiol. Rev.*, 1999, 12(1):147-179

# UNITED STATES PATENT AND TRADEMARK OFFICE

# CERTIFICATE OF CORRECTION

PATENT NO. : 7,338,794 Page 1 of 2

APPLICATION NO.: 10/681,540

DATED : March 4, 2008

INVENTORS : Frank H. Gaertner, Stacey Finstad Lee, Robert Shutter

It is certified that errors appear in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

Column 4,
Line 8, "2α/GROP" should read --2α/GROβ--.

Column 11,
Lines 17-18, "1'-16, 11-18" should read --11-16, 11-18--.

Column 23,
Line 17, "MVP(T" should read --MVP®--.

Column 24,
Line 48, "$\geqq 10^{10.6}$" should read --$\geq 10^{10.6}$--.

Column 25,
Line 20, "400%1" should read --400μ1--.

Column 28,
Lines 13-14, "bovine tracheitis" should read --bovine rhino tracheitis--.

Column 32,
Table 8, MCP-1/MCAF reference, Function column,
"aguments neutrophil" should read --augments neutrophil--.

MAILING ADDRESS OF SENDER:
Saliwanchik, Lloyd & Saliwanchik
P.O. Box 142950
Gainesville, FL 32614-2950

# UNITED STATES PATENT AND TRADEMARK OFFICE

# CERTIFICATE OF CORRECTION

PATENT NO. : 7,338,794 Page 2 of 2

APPLICATION NO.: 10/681,540

DATED : March 4, 2008

INVENTORS : Frank H. Gaertner, Stacey Finstad Lee, Robert Shutter

It is certified that errors appear in the above-identified patent and that said Letters Patent is hereby corrected as shown below:

Column 32,
Table 8, MIP-1β Reference, Function column,
"moncytes, densritic" should read --monocytes, dendritic--.

Column 37,
Lines 58-59, "gene encodes" should read --gene that encodes--.

Column 38,
Lines 60-61, "those cytokines or chemokines" should read --a cytokine or chemokine as--.

Column 39,
Line 21, "lipopolysaceharide" should read --lipopolysaccharide--.

MAILING ADDRESS OF SENDER:
Saliwanchik, Lloyd & Saliwanchik
P.O. Box 142950
Gainesville, FL 32614-2950